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<b>(54) Title:</b> METHOD AND APPARATUS FOR MEASURING LOCALLY AND SUPERFICIALLY THE SCATTERING AND ABSORPTION PROPERTIES OF TURBID MEDIA		
<b>(57) Abstract</b> <p>We present a method and apparatus for local and superficial measurement of the optical properties of turbid media. The depth probed is on the order of 1 transport mean free path of the photon. The absorption coefficient, reduced scattering coefficient and the phase function parameter <math>\gamma=(1-g_2)/(1-g_1)</math> are optical parameters computed from a single measurement of the spatially resolved reflectance close to the source. Images of superficial structures of the medium can be obtained by performing multi-site measurements. An important application of this technique is the characterization of biological tissues, for example for medical diagnostic purposes. Measurements on biological tissues can be achieved using a probe of diameter less than 2mm, and the average volume probed is on the order of 1 mm<sup>3</sup>. Separate images of absorption and tissue structure can be achieved with a resolution of approximately one transport mean free path of the considered tissue.</p> <div data-bbox="833 1207 1364 1963"><p>The diagram illustrates the experimental setup for measuring the reflectance of a turbid medium. A source and a detector are positioned on a horizontal surface, separated by a radial distance <math>\rho</math>. The source emits light into the turbid media at an angle <math>\omega_{source}</math>, and the detector receives light at an angle <math>\omega_{detector}</math>. Below the diagram, a graph shows the reflectance <math>R(\rho)</math> as a function of the radial distance <math>\rho</math>. The curve starts at a high value for small <math>\rho</math> and decreases as <math>\rho</math> increases. A text box next to the graph states: <math>R(\rho)</math> depends on <math>\mu_s, \mu_a</math> and <math>p(\theta)</math>.</p></div>		

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# METHOD AND APPARATUS FOR MEASURING LOCALLY AND SUPERFICIALLY THE SCATTERING AND ABSORPTION PROPERTIES OF TURBID MEDIA

## 5 1. Field of the invention

The present invention relates to a method and an apparatus to quantify the optical scattering and absorption properties of a turbid media. More precisely the present invention relates to a non-invasive measurement, over a small area of the sample surface. Local and superficial characterization of biological tissues in vivo is a major application of this invention.

## 2. Related Background Art.

Different techniques have already been proposed to quantitatively determine the absorption and reduced scattering coefficients of turbid media<sup>1</sup>. Most of the non-invasive methods are based on the measurement of spatially and/or temporally-resolved reflectance. The principle is as follows: the turbid medium is illuminated by a light source. The backscattered light is measured by one or several detectors. Different types of measurements are possible, depending on the time-dependence of the illuminating source: steady-state (continuous source), time-domain (short pulsed source) or frequency domain (amplitude modulated source). The present invention relates to the case of steady-state measurements performed at different distance  $\rho$  between the source and the detectors. However, the technique presented here can be complemented by time- or frequency-domain measurements. The range of  $\rho$  values is an important point to consider, when comparing different methods based on the measurement of the reflectance. First, the probed volume of the turbid

medium is related to the source-detector separation  $\rho$ . The larger the source-detector separation, the deeper the average depth probed. Second, depending on the range of  $\rho$ , different mathematical processing must be used to obtain the optical properties from the raw data.

At least two cases must be distinguished.

5 1) The first case corresponds to source-detector separations larger than several transport mean free paths. For typical biological tissue optical properties<sup>2</sup>, this case correspond to source-detector separation of larger than 2 mm. An analytical form of the reflectance can be obtained from the diffusion equation, if the absorption coefficient  $\mu_a$  is sufficiently lower than the scattering coefficient  $\mu_s$  (typically at least ten times). In such a case, the relevant optical properties are the refractive index, the absorption coefficient and the reduced  
10 scattering coefficient. The average depth of probing is on the same order  $\rho$  than the source-detector separation.

Such methods have been already published, and are the object of patents (Ref.1, Patent 5,517,987 Tsuchiya, Patent 5,676,142 Miwa et al.).

15 2) The second case corresponds to source-detector separations close to one transport mean free path. For biological tissues<sup>2</sup>, such source-detector separations correspond typically to distances from 0.1 to 2 mm. The average depth probed is on the order of 1 mm. Such small source-detector separations enable the measurement of the optical properties of a small tissue volume.

20 In this case, light propagation can be modeled using Monte Carlo simulations. In contrast to the previous case 1), it was discovered by the inventors that not only the first moment of the phase function must be considered, but also the second moment. More precisely, the relevant optical properties are the refractive index, the absorption, the reduced scattering

coefficient and the parameter  $\gamma=(1-g_2)/(1-g_1)$ , that takes into account the first two moments of the phase function, denoted  $g_1$  and  $g_2$  respectively.

Different methods have been proposed for local characterization of turbid media, and in particular of biological tissues. Nevertheless none allows for the simultaneous determination of the absorption, the reduced scattering coefficient and the parameter  $\gamma$ . Wang et al. (Patent 5,630,423) proposed a method for the determination of the reduced scattering coefficient only, using an optical beam of oblique incidence. Moreover, their analysis does not include the effect of the phase function. Kessler et al. (Patents 5,284,137 and 5,645,0619) proposed a method for measuring the local dye concentration and scattering parameters in animal and human tissues, based on spatial and spectral measurements. However, their methods do not enable the simultaneous determination of the absorption coefficient, reduced scattering coefficient and the parameter  $\gamma$ .

In the present invention we present a method and apparatus for the measurement of the absorption coefficient, the reduced scattering coefficient and the said phase function parameter  $\gamma$ , from the spatially-resolved reflectance data at short source detector separation. These three parameters, which can be measured at different wavelengths, enable us to characterize turbid media, such as biological tissues. Our method is based on a theoretical study of the reflectance at short source-detector separations, that we performed with Monte Carlo simulations. Other publications may be of concern:

1. See "Welch, A. J.; van Gemert, M. J. C. Optical Thermal Response of Laser Irradiated Tissue; Plenum publishing Corp., New York, 1995", and references therein.
2. W.-F. Cheong, S.A. Prahl, and A.J. Welch, "A Review of the Optical Properties of Biological Tissues," IEEE J. Quantum Electron. 26, 2166-2185 (1990).

### BRIEF DESCRIPTION OF THE DRAWINGS

Fig.1. Description of the principle of spatially-resolved reflectance measurement.

Fig.2. Probability density function of the mean depth of scattering events per detected photons.

Fig.3. Examples of reflectance obtained with different phase functions. Case of matched refractive index ( $n=1.0$ ).

Fig.4. Monte Carlo simulations and fits on the form  $R(\rho) \equiv [A(\rho, \mu_s', \gamma) + \mu_s' B(\mu_a)]^2$ . Case of constant  $\gamma=1.9$  and  $\mu_s'=1 \text{ mm}^{-1}$ , numerical aperture of the source and detectors = 0.37.  $A = 0.0647\rho^{0.324}\exp(-0.161\rho)$ ,  $B = 0.18653 \mu_a - 0.8466 \mu_a^2 + 1.836\mu_a^3$

Fig.5.a. Basic description of the apparatus: Case of measurement with an optical fiber probe

Fig. 5.b. Basic description of the apparatus: Particular embodiment where the probe is composed of  $n$  detecting fibers (typically 6). A broadband or white light source is used for excitation and the retrodiffused light is collected by the  $n$  detecting fibers and dispersed in parallel in a spectrograph, before being detected on a 2D CCD camera and transmitted numerically to a PC for  $\gamma, \mu_s', \mu_a$  determination at each wavelength  $\lambda$ .

Fig. 5.c. Basic description of the apparatus. Case of measurement with sources and detectors directly in contact with the turbid medium.

Fig. 5.d. Basic description of the apparatus. Case of non-contact measurements with a 1D or 2D detector, coupled with an imaging device. Scanning systems 1 and 2 are optional, they may be distinct from one another or confounded in a single scanner.

Fig. 5.e Basic description of the apparatus. Case of non-contact measurements with source and detection beams deflected by one or two scanning devices which may be distinct from one another or confounded.

Fig.6. Examples of the sample side of the fiber optical probe.

6a. simple arrangement for a single measurement.

6b. arrangement for symmetrical measurements.

6c. Arrangement for multiple measurements.

6d. Arrangement for multiple measurement, using an multicore fiber, or an optical fiber bundle.

5 6e. Arrangement for symmetrical measurements where detecting fibers are arranged on an elliptic path so as giving a regular spacing from the illuminating fibers to the detecting fibers, while maximizing the collecting surface of the fibers. Additional fibers F can be put in the center of the probe or around the probe for other light channels: collecting fibers for fluorescence or Raman scattering for example.

10 Fig.7.Example of an measured the spatially-resolved reflectance with the probe shown in Fig.6a. The measurement, performed on a microsphere suspension, is superimposed to a Monte Carlo simulation. The optical properties, computed from published water optical properties ( $\mu_a$ ) and Mie theory ( $\mu_s'$ ,  $\gamma$ ) are:  $n=1.33$ ,  $\mu_a=0.0004 \text{ mm}^{-1}$ ,  $\mu_s'=1.0 \text{ mm}^{-1}$ ,  $\gamma=2.2$ . The calibration was performed with a siloxane sample of known optical properties.

15 Fig.8.Relation between the parameters  $R(\rho=1 \text{ mm})$  and  $|\partial \rho \ln R(\rho=1 \text{ mm})|$  and the optical coefficients  $\mu_s'$  and  $\mu_a$ . Case of  $\gamma=1.5$  and  $1.9$ . Probe of refractive index =  $1.5$ , sample of refractive index =  $1.4$ , optical fibers diameter =  $200 \mu\text{m}$ ,  $\text{NA}=0.37$  (source and collection).

Fig.9.Plot of the parameter  $\frac{\partial}{\partial \rho} \sqrt{R}$  for different  $\gamma$  values ( $1.0, 1.8, 2.5$ ), different reduced albedo  $a'$  ( $1, 0.95, 0.9$ ) and for fixed  $\rho=1 \text{ mm}$ . Mismatched refractive index  $n=1.4$ .

Fig.10Plot of  $\sqrt{R(a'=1)} - \sqrt{R(a')}$  for different  $\gamma$  values and reduced albedo  $a'$ , and for fixed  $\rho=1 \text{ mm}$ . Mismatched refractive index  $n=1.4$ .

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## DETAILED DESCRIPTION OF THE INVENTION

### 1. Definitions

The concept of spatially resolved reflectance is illustrated in Fig.1. Consider light impinging on a turbid medium (through air or through a light guide), in a given solid angle  $\omega_{\text{source}}$ . The spatially resolved reflectance  $R(\rho)$  is defined by the backscattered light power in a given solid angle  $\omega_{\text{detector}}$  at a distance  $\rho$  from the source, per unit area and normalized by the source power. The distance  $\rho$  is referred as the source-detector separation (also in the case when light guides or imaging devices are used for the illumination and for the collection of the backscattered light).

$R(\rho)$  depends on the optical properties of the turbid medium, defined below. Note that  $R(\rho)$  depends also on the source and detectors characteristics, i.e numerical aperture and sizes.

It is commonly admitted that the fundamental optical properties of a turbid medium is determined by the average index of refraction  $n$  of the medium, the absorption coefficient  $\mu_a$ , the scattering coefficient  $\mu_s$ , and the phase function  $p(\theta)$  where  $\theta$  is the scattering angle. The absorption coefficient  $\mu_a$  [ $\text{mm}^{-1}$ ] is defined as the probability of absorption per unit infinitesimal pathlength. The scattering coefficient  $\mu_s$  [ $\text{mm}^{-1}$ ] is the scattering probability per unit infinitesimal pathlength. The phase function  $p(\theta)$  is the density probability function for the scattering angle  $\theta$ . The phase function is normalized as follows:

$$1 = 2\pi \int_0^\pi p(\theta) \sin\theta d\theta. \quad (1)$$

The  $n^{\text{th}}$  order moment  $g_n$  of the phase function is defined as:

$$g_n = 2\pi \int_0^\pi P_n(\theta) p(\theta) \sin\theta d\theta \quad (2)$$



where  $P_n(\theta)$  is the Legendre polynomial of order  $n$ . The first moment of the phase function is also called the anisotropy factor, and is often simply noted  $g$  ( $=g_1$ ). It represents the mean cosine of the scattering angle. The reduced scattering coefficient  $\mu_s'$  is defined as:

$$\mu_s' = \mu_s(1-g_1) \quad (3)$$

5 The transport mean free path (or reduced mean free path)  $mfp'$  is defined as:

$$mfp' = (\mu_s' + \mu_a)^{-1} \quad (4)$$

The reduced albedo  $a'$  is the ratio:

$$a' = \mu_s' / (\mu_s' + \mu_a) \quad (5)$$

10 As a result of the present invention described in more details in the next section, it is also necessary to define another phase function parameter called  $\gamma$ :

$$\gamma = (1-g_2)/(1-g_1) \quad (6)$$

All the parameters listed above are referred as optical properties. They are wavelength dependent, and can vary in space and time.

15

## 2. Reflectance measurements at distances close to one transport mean free path

The methods described in the Patent 5,517,987 (Tsuchiya) are based on measurements with large range of source-detector separations, typically from 1  $mfp'$  to 10  $mfp'$ . In such cases, the volume probed is on the order of  $10-1000 (mfp')^3$ . In contrast with such a large  
20 scale investigation, the present invention is directed to a novel approach where the volume probed is much smaller, on the order of  $1 (mfp')^3$ . This is achieved by using only small source detector separations, typically from 0.1 to 2  $mfp'$ . The lateral dimension of probing is limited to this range of distances.

Note that the present invention can be complemented by large source-detector separation measurements, if the optical properties of both superficial and deep parts of a turbid medium must be determined.

A model of photon migration in tissue was necessary to predict the relationship between the measured reflectance and the optical properties. Analytical solutions from the diffusion equation are not appropriate in our case because we are interested in the reflectance close to the source, at distances comparable to the transport mean free path [mfp']. We have performed Monte Carlo simulations to predict the measured reflectance of an homogeneous semi-infinite turbid media.

The exact diameter of the illuminating and collecting fibers, as well as their numerical apertures, have been taken into account in the simulations. The mismatch of index of refraction at the surface of the medium have been also taken into account in the simulations, by using the Fresnel law for each photon reaching the surface.

This is also one result of this simulation to compute the average depth of probing, illustrated in Fig.2. It is demonstrated that only the superficial part of the turbid medium is probed if small source-detector separations are used. For this, we determined the depth below the surface of each scattering event in the simulation. With this information we determined the average depth of all the scattering events for each detected photon, which we present as a probability density function in Fig.2. This figure shows that the average depth of scattering is approximately around 1 mfp'. Moreover it showed that the part located below 2 mfp' were not playing a significant role in the measured signal (for  $\rho < 1.5$  mfp').

The spatially resolved reflectance  $R(\rho)$ , with short source-detector separations is more

complex than in the case of large source-detector separations. Indeed, for small source detector-separation conditions, the inverse problem, i.e. calculating the localized absorption and reduced scattering coefficients, is sensitive to the scattering phase function. It is part of the present invention to have shown, from Monte Carlo simulations, that only the first and second moments of the phase function can be taken into account. Moreover, it was established that the influence of these two moments are not independent. Indeed, it is a merit of this invention to demonstrate that only one parameter  $\gamma = (1 - g_2)/(1 - g_1)$ , which depends on the first and second moments of the phase function can be used to characterize accurately the reflectance at short source-detector separation.

Fig.3 illustrates the fact that the parameter  $\gamma$  is the only predominant parameter of the phase function that must be taken into account (and not  $g_1$  as frequently mentioned in literature). Different reflectance curves, obtained from Monte Carlo simulations are shown in Fig.3. Four different phase functions were used for the simulations. Three phase functions are characterized by  $\gamma = 1.25$ , but different  $g_1$  and  $g_2$  values. Fig.3 shows that almost identical reflectance curve are obtained for distances  $\rho\mu_s' > 0.3$ , if the  $g_1$  and  $g_2$  are varied in such a way that the parameter  $\gamma$  stays constant ( $\gamma = 1.25$ ). For comparison, the reflectance computed with a different  $g$  value is also presented ( $\gamma = 2.25$ ). It appears then clearly that the parameter  $g$  plays a significant role in the reflectance.

The parameter  $\gamma$  depends on the characteristics of the set of scatterers inside the turbid medium: shape, distribution of sizes and refractive index.

The possibility of determining this parameter  $\gamma$  is an important part of this invention, and has never been reported earlier. The determination of  $\gamma$  can be related to the microscopic structure of the sample, since  $\gamma$  is related to the sizes and refractive indexes of the scatters.

In most cases, the average refractive indices  $n$  and the source and detectors characteristics are fixed and known. In such case, only three parameters are determined from measurements at short source-detector separation:  $\mu_a$ ,  $\mu_s'$  and  $\gamma$ .

It is important to note that the parameter  $\gamma$ , cannot be estimated if large source detector separations are used. Indeed, the reflectance is only sensitive to  $\gamma$  at short source-detector separation.

It was also derived from Monte Carlo simulations that the reflectance  $R(\rho)$ , for  $\rho \approx 1 \text{ mfp}'$ , can be reasonably well approximated by the expression:

$$R(\rho) \equiv [A(\rho, \mu_s', \gamma) + B(\mu_a, \mu_s')]^2 \quad (7)$$

In many cases, we found that Equ.(7) can also be written as:

$$R(\rho) \equiv [A(\rho, \mu_s', \gamma) + \mu_s' B(\mu_a)]^2 \quad (8)$$

it is important for the description of the invention to evidence the fact that the function  $A$  depends on  $\rho$ , the scattering properties (i.e.  $\mu_s'$  and  $\gamma$ ) but not on  $\mu_a$ . In contrast, the function  $B$  depends on  $\mu_a$  and  $\mu_s'$  but neither on  $\gamma$  nor  $\rho$ . An example of Equ.(8) is shown in Fig.4.

This formulation is of great help to solve the inverse problem, as described in section 5.3.

### 3. Apparatus

In the first embodiment, The apparatus can be divided in three parts, described in Fig.5.a.

The first part is the illuminating system. Any light source can be used. For examples: a) white sources, such as halogen or xenon lights, metal halides or fluorescent or phosphorescent sources. b) sources such as lasers, laser diodes, optical fiber lasers, light emitting

diodes or superluminescent diodes. c) the sources described in point a) and b) where monochromators, filters or interference filters are added to select a given set of wavelengths.

In the first preferred embodiment, The light power is conducted to the investigated sample by the probe, which is the second part of the apparatus. The probe is preferably made of optical fibers, to illuminate and to collect the backscattered light. But GRIN rods or other type of light pipes can also be used. Different possible arrangements of optical fibers are illustrated in Fig.6. Two different modes of measurements can be chosen. First, one fiber is used to illuminate the sample and at least two other fibers are used to collect the backscattered light at two different distances. Second, one fiber is used to collect the light and at least two other fibers, located at two different distances from the first one, are used to illuminate sequentially the sample.

The arrangement of the different fibers can be replaced by any imaging system or image guide, such as multicore optical fibers, or optical fiber bundle.

The light collected by the probe is analyzed by the detection unit, which is the third part of the apparatus. If wide spectral light sources are used (such as halogen or xenon lights), a spectrograph can be put between the probe and the detector to get wavelength dependence of the backscattered signal (either in the source or detection unit). Any types of detectors can be used. For example, photodiodes, avalanche photodiodes or photomultipliers can be assigned to each collecting fibers. Simultaneous detection of each collecting fiber can also be achieved using linear or two-dimensional detectors such as Charge-Coupled Detectors (1D or 2D), intensified CCD or array of photodiodes.

Aparticular embodiment is described in Fig. 5.b. : the probe is composed of n detecting

fibers (typically 6). A broadband or white light source is used for excitation and the retro-diffused light is collected by the  $n$  detecting fibers and dispersed in parallel in a spectrograph, before being detected by a 2D CCD camera and transmitted numerically to a PC for  $\gamma$ ,  $\mu_s'$ ,  $\mu_a$  determination at each wavelength  $\lambda$ . This particular embodiment has the advantage of yielding the wavelength dependance of  $\gamma$ ,  $\mu_s'$ ,  $\mu_a$  from a single measurement.

A second type of embodiment is presented on Fig. 5.c. The difference with embodiments presented in Fig. 5.a. and Fig. 5.b. is that optionally no optical fibers, light pipes or grin rods is used. The light source unit is directly in contact with the turbid medium, as well as the detector unit. Collimating optics, microoptics or imaging optics (DOE for example) can be put between the turbid medium and the actual light sources and detectors. The different type of sources and detectors cited in example for the first embodiment can be used for the second type of embodiments. Hybrid design, such as arrangements involving both direct contacts sensors or detectors and fibers, light pipes or grin rods are also included in the present embodiment.

The third embodiment is described in Fig. 5.d. and Fig. 5.e. The only difference with the first and second embodiment is that no contact measurements are performed. A collimating system allow for a point-like illumination on the turbid medium. An imaging system enables the measurement of the spatial distribution of the reflectance. The detectors can be either an array (1D or 2D) of detectors (Fig. 5.d) with optional scanning 1 and 2 which can operate separately or can be confounded in a single scanner, or a single detector (Fig. 5.e). In this last case, an scanning device is used to obtain the spatially-resolved reflectance. A fiber bundle, multicore fiber or relay optics (grin rod or multiple lenses) can be put between the focal point of the imaging system and the detector(s). The different type of

sources and detectors cited in example for the first embodiment can be used for the third embodiment.

Spatial images of the parameters  $\mu_a$ ,  $\mu_s$  and  $\gamma$  can be obtained by a series of measurements at different locations, that we call multi-site measurements. Some mechanical or optical scanning device can be used for this purpose. The resolution of such images is on the order of the mean source-detector separation used for a single site measurement. All three embodiments can be expanded to perform multi-site measurements, by duplicating and/or multiplexing the illuminating or measuring devices. For example, the optical fiber probes shown in Fig.6, can be duplicated and put side by side. The scanning system described in the third embodiment can also be expanded to perform multi-site measurements.

#### 4. Normalization and calibration

The differences of transmission between each fiber are corrected by performing a measurement on a turbid phantom illuminated uniformly.

The background light, measured with the light source turned off, must be subtracted from the signal.

In order to perform absolute intensity measurements, a calibration is performed on a turbid medium of known optical properties. Examples of such a medium are: 1) solid or liquid turbid medium which properties have been measured by other techniques, or reported in the literature 2) water suspension of microsphere of known size distribution and refractive index. Absorbing dye may be added to the suspension. In case 2) the scattering properties are calculated from Mie theory, and the absorption coefficient is assumed to be equal to the water absorption coefficient, if no absorbing dye is added. If an absorbing dye is used,

the absorption coefficient can be measured by a spectrophotometer, before mixing the solution with any scattering materials.

A Monte Carlo simulation is performed with the optical properties of the calibration sample. The simulation is then divided by the experimental reflectance performed on the calibration sample. The result, that must be independent of the source-detector separation, is defined as the calibration factor. Each new measurement is multiplied by the calibration factor.

An example of a measurement, after calibration, is shown in Fig.7. The measurement was performed with the apparatus described in the first embodiment, with a probe similar to the one described in Fig.6.a. A prior calibration was performed on a solid turbid medium, which optical properties were measured by another technique (frequency domain measurement, cf. Ref1). The sample was a water suspension of polystyrene microsphere of 1  $\mu\text{m}$  diameter. The measurement is compared to a Monte Carlo simulation performed with the scattering properties calculated from Mie theory and absorption coefficient of water. Fig.7 shows an excellent agreement between the measurement and the simulation, which confirm the validity of our simulation model, experimental measurement and calibration.

## 5. Signal processing

### 5.1. Control of the homogeneity of the area probed

Artifacts during a measurement, for example due to bad contact between the probe and the sample, or heterogeneity of the sample, can be detected by the following optional procedure. Two illuminating fibers are disposed symmetrically in regard to the collecting fibers (see Fig.6b). If the sample is homogeneous, the reflectance curve should be identical with



either illuminating fiber. Therefore, heterogeneity of the investigated region or obstructions beneath the fibers are detected by comparing the two curves. If the two curves are sufficiently close, the measurement is validated and the average of the two curves is calculated.

## 5 5.2. Smoothing procedure and computation of the derivative of the curve.

Functions in the form  $m_1 \rho^{m_2} \exp(m_3 \rho)$  were always found to fit well Monte carlo simulations for a restricted range of distances. Therefore, a smoothing of the experimental reflectance  $R(\rho)$  can be processed by fitting  $R(\rho)$  to  $m_1 \rho^{m_2} \exp(m_3 \rho)$ .

10 The determination of the slope of the logarithm  $\frac{\partial}{\partial \rho}(\ln R(\rho, \mu_s', a', \gamma))$  is also derived from this fit, using the following formula:

$$\frac{\partial}{\partial \rho}(\ln R(\rho, \mu_s', a', \gamma)) = \frac{m_1}{\rho} + m_2. \quad (9)$$

The slope of the square root of  $R(\rho)$  can also be derived (the use of these slopes are described below in section 5.3):

$$15 \quad \frac{\partial}{\partial \rho} \sqrt{R(\rho, \mu_s', \mu_a, \gamma)} = \left( \frac{m_1}{\rho} + m_2 \right) \left( \sqrt{m_1} \rho^{\frac{m_2}{2}} \exp\left(\frac{m_3 \rho}{2}\right) \right) \quad (10)$$

## 5.3. Inverse problem

We developed different methods to solve the inverse problem, which consists in extracting optical coefficients from the reflectance data.

### 20 Method 1.

Monte Carlo simulations show that the measurements of the reflectance intensity  $R(\rho)$  and the slope of  $\ln R(\rho)$  (denoted  $\partial \rho \ln R$ ), determined at a fixed distance  $\rho$ , can be used to derive  $\mu_s'$  and  $\mu_a$  for a given  $\gamma$  value. Fig.8 shows graphically the relationship between  $\mu_s'$

and  $\mu_a$  and the two parameters  $R(\rho=1 \text{ mm})$  and  $|\partial \rho \ln R(\rho=1 \text{ mm})|$  in the case of for  $\rho=1 \text{ mm}$ ,  $\gamma = 1.5$  and  $1.9$ . Note that any other choice of  $\rho$  is possible, as long as  $\rho$  is on the order of  $1 \text{ mfp}$ . We see in Fig.8 that  $\mu_s'$  and  $\mu_a$  can not be determined uniquely if  $\gamma$  is unknown. Further insight for optimizing the inversion strategy is provided by three additional features of Fig.8.

First, the determination of  $\mu_s'$  is only weakly influenced by  $\gamma$ . Indeed in Fig.8 the errors induced by error in  $\gamma$  are typically 10% for  $\mu_s'$  around  $1 \text{ mm}^{-1}$ . Second, although absolute determination of  $\mu_a$  is not possible when  $\gamma$  is not precisely known, relative absorption changes can be still evaluated. Third, the indetermination of the parameter  $\gamma$  may be resolved by the values of  $R(\rho)$  and/or  $|\partial \rho \ln R(\rho)|$  at other distances. Therefore the following procedure can be used:

(1) determination of  $\mu_s'$  and  $\mu_a$  from  $R(\rho=1 \text{ mm})$  and  $|\partial \rho \ln R(\rho=1 \text{ mm})|$  for a set of values  $\gamma$ . For example:  $\gamma=1.0, 1.1, 1.2, \dots, 2.5$ .

(2) simulations with the different sets of  $\mu_s'$  and  $\mu_a$  obtained from point 1)

(3) comparison between the simulations and the reflectance profile for distances  $0.3 < \rho < 2 \text{ mm}$ .

This last step allows us to determine the correct values of  $\gamma$ ,  $\mu_s'$  and  $\mu_a$ . Points 1 to 3 can be done iteratively to evaluate  $\gamma$  more precisely, using a finer discrimination of  $\gamma$  values.

This method described by the iteration of points 1) to 3) corresponds to finding a simulation that fit closely a measured curve  $R(\rho)$ . This can also be achieved by directly comparing the measured curve  $R(\rho)$ , with a set of simulations, and finding the simulation that minimize the square of the differences between the measured and simulated curves.

Note also that the use of the reflectance intensity  $R(\rho)$  and the slope of  $\ln R(\rho)$  is equivalent

to the use of two intensities  $R(\rho_1)$  and  $R(\rho_2)$ , measured at two close distances  $\rho_1$  and  $\rho_2$ , respectively.

Method 2.

5 The inverse problem can also be performed, considering properties of  $R(\rho)$  expressed by Equ.(7).

Indeed, it can be derived from Equ.(7) that the quantity  $\frac{\partial}{\partial \rho} \sqrt{R(\rho, \mu_s', \mu_a, \gamma)}$  does not depend on the absorption coefficient  $\mu_a$ :

$$\frac{\partial}{\partial \rho} \sqrt{R(\rho, \mu_s', \mu_a, \gamma)} = 2 \frac{\partial A}{\partial \rho}(\rho, \mu_s', \gamma) \quad (11)$$

10 This property is confirmed in Fig.9, where the quantity  $\frac{\partial}{\partial \rho} \sqrt{R(\rho, \mu_s', \gamma)}$ , evaluated at  $\rho=1$  mm, is plotted as a function of  $\mu_s'$  for  $\gamma=1, 1.9$  and  $2.5$  and reduced albedo  $a'=1, 0.95$  and  $0.9$  (note that any other choice of  $\rho$  is possible, as long as  $\rho$  is of the order of 1 mfp').

In Fig.9, the parameter  $\frac{\partial}{\partial \rho} \sqrt{R(\rho, \mu_s', \gamma)}$  clearly depends on  $\mu_s'$  and  $\gamma$ . In contrast, the dependence on  $a'$  is almost negligible.

15 Therefore,  $\gamma$  and  $\mu_s'$  can be derived from the parameter  $\frac{\partial}{\partial \rho} \sqrt{R(\rho)}$ , calculated from the experimental reflectance  $R(\rho)$  (see section 5.2). Simultaneous determination of  $\gamma$  and  $\mu_s'$  require values of  $\frac{\partial}{\partial \rho} \sqrt{R(\rho)}$  at different distances  $\rho$  (at least two). If one of the parameters  $\gamma$  or  $\mu_s'$  is already known, the determination of the other one can be obtained from the value of  $\frac{\partial}{\partial \rho} \sqrt{R(\rho)}$  at a single distance.

20 Convenient analytical approximation of  $\frac{\partial}{\partial \rho} \sqrt{R(\rho)}$  can be obtained by fitting Monte Carlo results to polynomial functions. For example, in the case of fixed  $\gamma=1.9$ , we obtained:

$$\mu_s' = 0.9162 - 52.89x + 1795x^2 - 18155x^3 + 65428x^4 \quad (12)$$

where  $x = \frac{1}{2\partial\rho}(\sqrt{R(\rho)})$  at  $\rho=1$  mm.  $x$  is expressed in  $[\text{mm}^{-2}]$  and  $\mu_s'$  is expressed in  $[\text{mm}^{-1}]$ .

This results is valid for a probe of refractive index of 1.5 and a sample of refractive index of 1.4, optical fibers of diameter 200  $\mu\text{m}$ , NA=0.37 (source and collection).

Once  $\mu_s'$  and  $\gamma$  are calculated from the procedure explained above,  $\mu_a$  is calculated from the absolute value of  $R(\rho)$ , which highly depends on  $\mu_a$ . For given  $\mu_s'$ ,  $\gamma$  and  $\rho$  values, the dependence of reflectance on  $\mu_a$  is obtained by Monte Carlo results. From Equ.(7) we have:

$$\mu_a = h[\sqrt{R(\rho)} - f(\gamma, \mu_s')] \quad (13)$$

where  $f$  and  $h$  are functions given by Monte Carlo simulation. Particularly, they can be well approximated by polynomial functions. For example, for  $\gamma=1.9$ , probe refractive index of 1.5, sample refractive index of 1.4, optical fibers of diameter 200  $\mu\text{m}$ , NA=0.37 (source and collection):

$$h = -0.002257 - 8.171y + 268.8y^2 \quad (14)$$

$$f = 0.01311 + 0.05184\mu_s' - 0.01974\mu_s'^2 + 0.003217\mu_s'^3 - 0.0001992\mu_s'^4 \quad (15)$$

where  $y = [\sqrt{R(\rho)} - f] [\text{mm}^{-1}]$ .  $\mu_a$  is expressed in  $[\text{mm}^{-1}]$ .

### Method 3

Equ.(7) also shows that relative measurements of  $\mu_a$ , i.e. variation of  $\mu_a$  from a known value  $\mu_{a0}$ , is possible by measuring the variation of the parameter  $\sqrt{R(\rho)}$ :

$$\begin{aligned} \sqrt{R(\rho, \mu_s', \gamma, \mu_{a0})} - \sqrt{R(\rho, \mu_s', \gamma, \mu_a)} \\ = B(\mu_{a0}, \mu_s') - B(\mu_a, \mu_s') \end{aligned} \quad (16)$$

This relation is illustrated in Fig.10 in the case of  $\mu_{s0}=0$ ,  $\rho=1$  mm,  $\gamma=1.0, 1.9$  and  $2.5$ , and for  $a'=1$  to  $0.83$ . Fig.10 confirms that the influence of  $\gamma$  is weak in the quantity  $\sqrt{R(\rho, \mu_s', \gamma, \mu_a=0)} - \sqrt{R(\rho, \mu_s', \gamma, \mu_a)}$ . For known  $\mu_s'$ , the function  $B(a')$  allows for the determination of a relative absorption change  $\Delta\mu_a = \mu_a - \mu_{s0}$ . Fig.10 illustrates the case  $\mu_{s0}=0$ , but any other value of  $\mu_{s0}$  is possible. The interesting point is that  $B(a')$  does not depend on the phase function.

## CLAIMS

1. A method for local and superficial (on the order of one transport mean free path) characterization of a turbid media using the following parameters:

- 1) the refractive index  $n$  of the turbid media
- 2) the absorption coefficient  $\mu_a$  of the turbid media
- 3) the reduced scattering coefficient  $\mu_s'$  of the turbid media
- 4) the phase function parameter  $\gamma = (1-g_2)/(1-g_1)$  of the turbid media, where  $g_1$  and  $g_2$  are the first two moments of the phase function.

and comprising the steps of:

-measuring the spatially-resolved reflectance  $R(\rho)$  or any quantity which allows for the indirect determination of the said spatially-resolved reflectance  $R(\rho)$ .

-mathematically processing  $R(\rho)$  to compute at least one of the said parameters ( $n$ ,  $\mu_a$ ,  $\mu_s'$ ,  $\gamma$ ) or the relative variations of these said parameters.

2. The method of claim 1, wherein said spatially resolved reflectance is measured by an optical fiber probe as described by the first embodiment in section 3, and described on fig 5. a.

3. The method of claim 1 and 2, wherein said spatially resolved reflectance is measured by an optical fiber probe composed of n fibers, as described in the particular embodiment in section 3, and described on fig 5.b, giving the wavelength dependance of at least one of the parameters ( $n, \mu_a, \mu_s', \gamma$ )

4. The method of claim 1, wherein said spatially resolved reflectance is measured by a device put directly in contact to the turbid media, as described by the second embodiment in section 3 and described on fig 5.c..

5. The method of claim 1, wherein said spatially resolved reflectance is measured by non contact detection unit, as described by the third embodiment in section 3 and described on fig 5.d.

6. The method of claims 1 to 5, wherein the following processing is performed:

- fit of the measured reflectance  $R(\rho)$  to the function:

$$m_1 \rho^{m_2} \exp(m_3 \rho) \quad (17)$$

This fit gives the values for the parameters  $m_1, m_2$  and  $m_3$ , assuming that the expression  $R = m_1 \rho^{m_2} \exp(m_3 \rho)$  gives a smooth function  $R(\rho)$ .

The slopes  $\frac{\partial}{\partial \rho} \sqrt{R(\rho)}$  and  $\frac{\partial}{\partial \rho} (\ln R(\rho))$ , or any mathematical combinations of these two latter quantities and  $R(\rho)$ , can be obtained directly from analytical functions using the parameters  $m_1, m_2, m_3$ , or by numerical procedures from the function  $R = m_1 \rho^{m_2} \exp(m_3 \rho)$ .

7. The method of claims 1 to 6, wherein the absorption coefficient  $\mu_a$ , the reduced scattering coefficient  $\mu_s'$  and the phase function parameter  $\gamma$  are determined on the basis of a fit of the measured spatially-resolved reflectance  $R(\gamma, \mu_s', \mu_a, \rho)$  to a set of Monte Carlo simulations, or functions approximating Monte Carlo simulations.

8. The method of claims 1 to 6, wherein the absorption coefficient  $\mu_a$ , the reduced scattering coefficient  $\mu_s'$  and the phase function parameter  $\gamma$  are determined on the basis of the following form of the reflectance:

$$R(\rho) = (A(\rho, \gamma, \mu_s') + B(\mu_a, \mu_s'))^2 \quad (18)$$

5 where the function  $A(\rho, \gamma, \mu_s')$  and  $B(\mu_a, \mu_s')$  depends also on the sources and detectors characteristics, and the refractive index of the sample.

9. The method of claims 1 to 6, wherein the reduced scattering coefficient  $\mu_s'$  and the phase function parameter  $\gamma$  are determined on the basis of the quantity  $\frac{\partial}{\partial \rho} \sqrt{R(\rho, \mu_s', \gamma)}$ , which depends weakly on the absorption coefficient  $\mu_a$  for  $0.3 < \rho \mu_s' < 5$ .

10 10. The method of claims 1 to 6, wherein the absorption coefficient  $\mu_a$  is determined based on the equation:

$$\mu_a = h[\sqrt{R(\rho)} - f(\gamma, \mu_s')] \quad (19)$$

where  $f$  and  $h$  are functions that approximate Monte Carlo simulations.

15 11. The method of claims 1 to 6, wherein variation of the absorption coefficient  $\mu_a$  from a known value  $\mu_{a0}$  is computed from the variation of the parameters  $\sqrt{R(\rho, \mu_s', \gamma, \mu_{a0})} - \sqrt{R(\rho, \mu_s', \gamma, \mu_a)}$ .

12. The method of claims 1 to 10, wherein said turbid medium is a biological medium.

20 13. The method of claims 1 to 11, wherein the measurement and processing is repeated at different location of the sample (multi-site measurements), to build images of one of the said parameters  $(n, \mu_a, \mu_s', \gamma)$ .

14. An apparatus for local and superficial characterization of a turbid media comprising a source, a detection unit, reference means, signal processing means, at least one source



connected to said source unit and at least one detector connected to said detection unit, the distance between the source and the detector being less than one transport mean free path.

15. The apparatuses of claim 14 characterized by the fact that the distance between the source and the detector varies from 0.1 to 2mm. for application to biological media and to turbid media similar to biological media.

16. The apparatus of claim 14 or 15 using the method of any claim 1 to 13.

17. The apparatus of claim 16 where the control of the homogeneity of the probed sample area is performed according to the procedure described in section 5.1.

18. The apparatus of claim 16 where the calibration and normalization procedures of section 4. are carried out.

# INTERNATIONAL SEARCH REPORT

International Application No.

PCT/CH 99/00476

## A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 G01N21/49

According to International Patent Classification (IPC) or to both national classification and IPC

## B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 G01N A61B

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	F. BEVILACQUA ET AL: "In vivo local determination of tissue optical properties" SPIE, EUROPEAN BIOMEDICAL OPTICS, BIOS EUROPE 97, vol. 3194, 1997, pages 262-268, XP000866481 page 267, last paragraph page 265, line 7 -page 266, line 10 ---	1,12,14, 15
A	US 5 452 723 A (WU JUN ET AL) 26 September 1995 (1995-09-26) column 12, line 43 - line 54 column 10, line 21 - line 33 column 6, line 44 -column 7, line 15 --- -/--	1,14



Further documents are listed in the continuation of box C.



Patent family members are listed in annex.

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Name and mailing address of the ISA

European Patent Office, P.B. 5818 Patentlaan 2  
NL - 2280 HV Rijswijk  
Tel. (+31-70) 340-2040, Tx. 31 651 epo nl,  
Fax: (+31-70) 340-3016

Authorized officer

Verdoodt, E

# INTERNATIONAL SEARCH REPORT

International Application No

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## C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	<p>P. MARQUET ET AL: "DETERMINATION OF REDUCED SCATTERING AND ABSORPTION COEFFICIENTS BY A SINGLE CHARGE-COUPLED-DEVICE ARRAY MEASUREMENT, PART I: comparison between experiments and simulations"</p> <p>OPTICAL ENGINEERING, vol. 34, no. 7, July 1995 (1995-07), pages 2055-2063, XP002129315</p> <p>abstract</p> <p>page 2057, left-hand column, line 1 -page 2058, left-hand column, line 6</p> <p style="text-align: center;">----</p>	1, 14
A	<p>D.R. WYMAN ET AL: "Similarity relations for the interaction parameters in radiation transport"</p> <p>APPLIED OPTICS, vol. 28, no. 24, 15 December 1989 (1989-12-15), pages 5243-5429, XP000086704</p> <p>page 5245, right-hand column, line 1 -page 5246, left-hand column, line 2</p> <p>abstract</p> <p style="text-align: center;">-----</p>	1

# INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/CH 99/00476

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 5452723	A	26-09-1995	NONE